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# Cereal Leaf Beetle Survey and Biocontrol Activities in Washington State, 2001

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#### **Background**

The cereal leaf beetle, *Oulema melanopus* (L.), is an exotic leaf-feeding pest of small grains such as oats, wheat, and barley. Originally from Europe, the cereal leaf beetle (CLB), was first detected in the United States in Michigan in 1962 (Castro et al. 1965). Since that time, the geographic range of CLB in the US has increased to most eastern states and many western states such as Idaho, Montana, Oregon, Wyoming, and Utah.

The first Washington State detection of cereal leaf beetle occurred in 1999 at four sites in Spokane County (Klaus et al.1999). Survey efforts were expanded the following year, resulting in detections at 13 additional sites in Lincoln, Pend Oreille, Spokane and Stevens counties (Hitchcox et al. 2000). While infestations of cereal leaf beetle have the potential to cause yield loss in small grains, populations in Washington State have not yet reached economically damaging levels. To date, the impacts of CLB on Washington agriculture have been mostly regulatory, triggered by quarantines in California and Canada.



CLB larvae and feeding damage on wheat.

Entomologists from the Washington State Department of Agriculture (WSDA), the U.S. Department of Agriculture (USDA) and Washington State University (WSU) are working cooperatively to study the distribution, biology, and control of the insect pest in the state. Fortunately, past work on the biological control of CLB has provided alternatives for long-term pest management. The WSDA is working with farmers to use selected natural enemies to suppress CLB populations below economic threshold levels. The results of the 2001 CLB survey, and the related biocontrol activities are reported herein.

#### **2001 Project Objectives**

- 1. Continue statewide CLB detection survey for regulatory purposes, with efforts placed on eastern Washington counties and selected western Washington counties.
- 2. Continue delimiting the geographical distribution of, and continue monitoring the density of, known CLB populations.
- 3. Continue biological control efforts through the maintenance of an insectary site and release of selected biocontrol agents.

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#### **PROJECT METHODS**

### 1. Detection Survey

Goals for the 2001 detection survey focused on sampling counties that 1) were not previously surveyed in eastern Washington by WSDA (Okanogan, Columbia, Franklin, Chelan, Yakima, Benton, and Ferry), and 2) were considered high priority western counties along the southern I-5 corridor (Lewis, Pierce, Thurston, Cowlitz, and King). The survey also included high priority counties previously surveyed (Adams, Clark, Douglas, Garfield, Grant, Kittitas, Klickitat, Lincoln, Pend Oreille, Spokane, Stevens, Walla Walla, Whitman). High priority counties were defined as areas with high economic risk, with suitable habitat, and adjacent to previously detected populations.

The western Cereal Leaf Beetle Working Group (CLBWG) serves as an informal scientific forum and regional partnership to aid in the exchange and redistribution of biological control agents. The group represents 10 western states/provinces, whose members include industry representatives, federal and state agricultural entomologists, and university researchers and extension agents. In 2001, members of the CLBWG met to review the survey methodology used for the detection of cereal leaf beetle. The group developed a list of survey guidelines and standardized methods for field sampling during CLB detection surveys.

Essentials of survey methods include both visual inspection and sweep netting. For survey purposes, a "sweep" was defined as one pass through the upper foliage with a 15" diameter sweep net. A "sample" was defined as 30 sweeps taken at a moderate walking pace 10-15 ft. inside the border of the field. Four samples were taken from each site, totaling 120 sweeps per site. The contents of each sample were visually inspected for life stages of CLB and all suspect specimens were retained for verification and reference. Guidelines advise surveying a minimum of 5 high priority fields per county, and a maximum of 20 sites per county, or until a detection is made. WSDA survey efforts concentrated on "high priority" sites which included commercial grain and grass fields such as oats, wheat, barley and rye. In areas with little grain production, sampling focused also on wild and cultivated grasses such as timothy, orchard grass, bluegrass, wild oats, corn, brome, and ryegrass.

#### 2. Delimitation Survey

A delimitation survey was conducted as a multi-year effort to monitor growth and movement of the localized CLB populations in the State. Because of limited resources, inspections were focused on fields adjacent to previously surveyed sites. Special attention was placed on potential avenues of natural and human-assisted dispersal such as river corridors, railway systems, and highways. Sampling methods for each site were identical to those used for the detection survey.

## 3. Biological Control

In 2000, the WSDA began a biological control program aimed at the long-term management of cereal leaf beetle. With the cooperation of a local farmer, the WSDA has established one CLB field insectary, located at Nine Mile Falls in Spokane County. The insectary site is located on approximately 600 acres of irrigated grain and grass farmland, bordered by forested slopes and waterfront. Later that same year, with the support of the USDA and

WSU, the WSDA conducted the initial release of the parasitic wasp species *Tetrastichus julis*. Renowned for its efficacious control of CLB, *T. julis* wasps locate young CLB larvae and deposit eggs into the host's body. After the eggs hatch, the emerging parasitic larvae begin to feed and develop slowly within the living host. The CLB larvae continue feeding and eventually move to the soil and build a pupal cell. After the pupal cell is constructed, the *T. julis* larvae grow rapidly, killing the CLB host. Some *T. julis* larvae will complete development immediately, emerge as wasps and disperse to attack new hosts. Another portion of the *T. julis* larvae will enter diapause and overwinter within the remaining host pupal cell, emerging as adults the following spring. (Gage and Haynes 1975, Stehr 1970).

In 2000 and 2001, Washington received shipments of the bioagent *T. julis* from university and federal cooperators in Utah<sup>i</sup> and Montana<sup>ii</sup>. During the first year, shipments were received and screened by the Northwest Biological Control Insectary/Quarantine (NWBIQ). Adult wasps were reared from the parasitized CLB and released at the Nine Mile Falls insectary site. In 2001, instead of rearing and releasing adult wasps, the parasitized CLB larvae were released directly at the Nine Mile Falls insectary site. This was done in order to 1) increase survivability of the parasitoid, 2) encourage synchronized emergence of *T. julis* wasps with local field conditions, and 3) make use of the diapausing bioagents that may be

dormant in the samples. To further facilitate rapid establishment of *T. julis*, a "conservation strip" strategy was initiated in 2001. The main purpose of this strategy is to reduce mortality of over-wintering *T. julis* caused by routine farm management practices. The conservation strips were defined as a 1-meter wide strip along the edge of a field, running parallel with the crop row. Each conservation strip was located in heavily infested areas of the farm, adjacent to winter and spring grains, and grass varieties. Shipments of parasitized CLB larvae were opened on site and releases were concentrated in these managed areas.



Parasitic wasp, <u>Tetrastichus julis</u> attacking a CLB larva at Nine Mile Falls insectary.

## Pre-release Survey

A "pre-release" study was conducted to monitor for naturally occurring larval parasitoids and to evaluate the success of the previous years biocontrol release. In June, prior to any 2001 bioagent releases, the WSDA and WSU personnel conducted a pre-release survey at the Nine Mile Falls field insectary by collecting CLB larvae from three winter wheat fields. Collected larvae were stored in 20% ethyl alcohol at 45°F for 1 week. Most larvae were sent directly to the USDA-APHIS-PPQ Niles Biological Control Laboratory (NBCL) for dissection and examination. Dissection techniques were also applied to screen for the presence of larval parasitic wasp larvae. A number of dissections were also completed by the WSDA.

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#### RESULTS and DISCUSSION

## 1. <u>Detection Survey</u>

Cereal leaf beetle adults were first observed in late March in Spokane County, with first eggs observed on May 23. The 2001 CLB survey season began on 25 April and ended 30 August. A total of 23 Washington counties were surveyed, which included five western counties and almost all counties east of the Cascade crest. As a result of the 2001 survey, CLB was positively detected in nine counties (Table 1). Five of those counties (Adams, Clark, Columbia, Franklin, and Grant) are reported as new county records for CLB. The remaining four counties (Spokane, Stevens, Pend Oreille and Lincoln) were reconfirmed positive in 2001.

Table 1: CLB Detection Survey summary, 2001						
County	# sites sampled	# positive sites	total # CLB found			
Adams	3	2	3			
Benton	10					
Clark	10	3	3			
Chelan	1					
Columbia	10	1	1			
Cowlitz	5					
Douglas	8					
Ferry	5					
Franklin	6	1	3			
Garfield	15					
Grant	6	2	5			
King	5					
Kittitas	20					
Klickitat	13					
Lewis	6					
Lincoln	2	1	1			
Okanogan	5					
Pend Oreille	1	1	1			
Spokane	3	3	61*			
Stevens	2	1	2			
Walla Walla	15					
Whitman	19					
Yakima	16					
* Dags not include NN	186	15				

<sup>\*</sup> Does not include NMF insectary site.

Evidence of CLB life stages (i.e., adults, larvae, eggs, and/or feeding damage) was detected at 15 of the 186 sites surveyed. Crop types represented at these positive sites include oats, wheat (spring and winter), triticale, barley, and mixed pasture grass. Detections were made at both irrigated and non-irrigated fields. Due to heightened survey protocol and limited staffing, not all 2001 survey goals were accomplished. Thurston and Pierce counties were not surveyed. Furthermore, survey efforts in many counties were below the targeted 20 fields.

The addition of positive counties could be a result of an increase in survey efforts over three years (See summary and Figure A). However, the rapidly expanding distribution of CLB is most likely attributed to the natural and human-aided dispersal of the pest, a trait commonly demonstrated in other states (Bai et al. 2001; Klaus 1992; NAPIS 2001).

## 2. Delimiting survey

Positive counties identified in 2000 (Spokane, Stevens, Lincoln and Pend Oreille) were also surveyed and confirmed positive in 2001. In Spokane County, a second-year detection was made in the Peone Prairie area, with larval density averaging 3 per ½ m, which suggests an established population. The discovery of CLB larvae at a second site in northcentral Spokane County suggests that populations are sustained and widespread in that area. In Lincoln, Pend Oreille and Stevens Counties, positive detections remain at low levels, typically resulting in the collection of 1-4 adult or larval specimens per site. The detection of CLB in Adams, Franklin, Grant and Columbia Counties suggests that separate infestations are occurring throughout the state. In Grant County, initial CLB observations by a WSU-NWBIQ entomologist reported beetle activity in the Stratford area east of Soap Lake in April 2002. In early May, WSDA confirmed the report resulting in detections of low numbers of CLB adults at two separate winter wheat fields near Stratford. These Grant County sites are geographically isolated from other previous detection sites in the state, the closest being in Lincoln County, approximately 70 miles away. Similar isolated occurrences were found in Adams, Franklin and Columbia Counties. While the specific routes of infestation are unknown, these new sites are located along or within 1 mile of the interstate or primary highways, which suggests human-aided movement of the insect.

## 3. Biocontrol

*Evaluation:* Prior to the 2001 release of new bioagents, 300 mid-late instar CLB larvae were hand collected from the Nine Mile Falls Field Insectary by WSU and WSDA staff. Larvae were dissected to monitor for *T. julis* parasitism. No parasitism was observed during this first-year evaluation.

Augmentation: Three separate shipments of parasitized CLB larvae were collected from insectaries in Montana and Utah. WSDA entomologists assisted in the field collection of biocontrol agents from Utah. Each shipment was released in Washington at the Nine Mile Falls Field Insectary in Spokane County. During 2001, approximately 7700 parasitized CLB larvae, with an estimated total of 9435 *Tetrastichus julis*, were released at the Nine Mile Falls field insectary (see Table 2). The two July shipments were released in the defined "conservation strip" areas. According to the grower, tilling was mostly suspended in these areas which should increase survivability of the overwintering parasitoids.

Table 2: Tetrastichus julis introductions to NMF insectary in 2001.						
Date released	Origin	# CLB larvae released	% Parasitism	Estimated # T. julis*		
20 June	Montana	3800	est. 35%	3990		
02 July	Utah	1900	86.7%	4845		
11 July	Montana	2000	10%	600		
TOTAL	-	7700	-	9435		

<sup>\*</sup> Estimate based on an average of 3 *T. julis* individuals per 1 CLB larva.

#### Conclusion

After three years of survey, CLB has been detected in a total of nine Washington counties. Cereal leaf beetle will most likely continue to expand its range in Washington State. This spread will probably be facilitated both by natural and human-aided movement. The state-wide detection survey will continue in 2002 targeting those counties not yet surveyed. Repeat surveys of previous detection sites will attempt to monitor known CLB populations and to identify new colonies that may have established. Supplemental releases of the biological control agent *Tetrastichus julis* are planned for the Nine Mile falls field insectary. The WSDA intends to set up additional field insectaries for the propagation of this and other species of biocontrol agents. The WSDA will continue cooperative work with growers, the USDA, WSU, and other western cooperators to study the distribution, biology, and natural enemies of the pest.

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### SUMMARY of ACTIVITY, 1999-2001

## **<u>Detection Survey</u>** (See Figure A)

Current positive counties reported (total number of positive sites):
Adams(2), Clark (3), Columbia (1), Franklin (1), Grant (2), Lincoln (3), Pend Oreille (2), Spokane (11), Stevens (4).

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Year	1999	2000	2001
Total counties surveyed	9	16	23
Total sites surveyed	135	200	186
Total positive counties	1	4	9
Total positive sites	4	14	15
duration	3 June - 28 July	27 Apr – 28 July	25 Apr - 30 Aug

## **Biological control**

- Implementation of "conservation strip" strategy at the Nine Mile Falls field insectary, to encourage establishment of *Tetrastichus julis* for control of CLB.
- Release of approximately 7700 parasitized CLB larvae at field insectary in 2001. Based on % parasitism, an estimated 9435 *T. julis* larvae were introduced. Total estimated number of *T. julis* introduced to insectary during both years: 10,135 larvae or adults.

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#### **Distribution / Content Note**

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